Diagnosis of Genital *Chlamydia trachomatis* Infections in Asymptomatic Males by Testing Urine by PCR

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An enzyme-linked immunosorbent assay (EIA) (MicroTrak; Syva) was compared with PCR (Amplicor; Roche) for detection of *Chlamydia trachomatis* in first-void urine (FVU) from 184 men attending a skin and venereal disease clinic. The prevalence of *C. trachomatis* in the population studied was 18.5%. Discrepant results between Syva EIA and Roche PCR were retested by using major outer membrane protein primer-based PCR. After retesting, the sensitivity, the specificity, and the positive and negative predictive values for the Syva EIA were 85.3, 100, 100, and 77.5%, respectively, and those for the Roche PCR 100, 100, 100, and 100%, respectively. It was concluded that PCR provides a highly sensitive and specific noninvasive screening method for genital chlamydial infection in asymptomatic men.

Detection of chlamydial antigen in sediments of first-void urine (FVU) from men is a feasible and accurate test for chlamydial urethritis (5, 13, 20). The use of FVU, especially for asymptomatic carriers, is more acceptable to males than urethral swabbing (4). The clinical and epidemiological implications associated with undetected genital chlamydial infections demand a rapid and accurate laboratory screening test (12, 21, 23).

Asymptomatic male carriers of *Chlamydia trachomatis* are of particular concern because the infection may be passed to female partners, resulting in pelvic inflammatory disease and its sequelae, i.e., ectopic pregnancy and infertility.

Our study was aimed at evaluating a PCR test for the detection of *C. trachomatis* in male FVU and comparing it with an enzyme immunosassay (EIA) test.

**MATERIALS AND METHODS**

**Study population.** FVU from asymptomatic males (*n* = 184) presenting at a skin and venereal disease clinic, for reasons other than venereal disease, was collected. Patients were supplied with 20-ml urine tubes and were asked to return after having collected their early-morning urine. The median age of the patients was 32 years.

**Specimen collection and preparation.** Fresh urine was divided into two aliquots of 10 ml each. One aliquot of urine was immediately centrifuged at 1,500 × g for 15 min, and the pellet was resuspended in 1 ml of Syva MicroTrak specimen treatment solution. The samples were kept at 4°C for ≤2 weeks before being investigated.

The other aliquot of urine was frozen at −20°C and kept for about 2 weeks until the PCR was done. Before the investigation, urine was thawed at room temperature and then heated for 30 min at 37°C to dissolve any precipitate. Then the specimens were vortexed (~1 min) and centrifuged at 1,500 × g for 10 min at room temperature. The supernatants were replaced with 2 ml of Roche Amplicor urine resuspension buffer and kept for 1 h at room temperature. After 2 ml of urine diluent had been added, the sample was left for 10 min at room temperature before amplification.

**Testing of urine.** EIA with the MicroTrak Chlamydia EIA was done by following the instructions of the manufacturer (Syva). PCR (Amplicor; Roche) based on plasmid primers was performed according to the manufacturer's instructions. The result was read with a Syva MicroTrak EIA autoreader at a wavelength of 450 nm.

Samples considered PCR positive were those having optical densities higher than 2.0 Δ₄₅₀ units. Optical densities for the negative samples were below 0.25 Δ₄₅₀ units.

**Analysis of discrepant results.** All samples giving discordant results were retested twice, by both plasmid-based PCR and EIA. Samples remaining positive only by plasmid-based PCR were sent to Roche Laboratories (Switzerland) to be retested with major outer membrane protein (MOMP) primer-based PCR.

**RESULTS**

One hundred eighty-four FVU samples from males were obtained for the EIA and PCR assay. Of the 184, 29 (15.8%) were positive by both EIA and PCR. Five FVU samples (2.7%) were positive for *C. trachomatis* by the plasmid-based PCR but negative by the Syva EIA. One hundred fifty EIA-negative FVU samples were also PCR negative.

Compared with EIA, plasmid primer-based PCR had a sensitivity, specificity, and positive and negative predictive values of 100, 98.6, 85.3, and 100%, respectively (Table 1). In light of these test results for the five samples, they were thought to have false-positive PCR results. In repeated plasmid primer-based PCR and in MOMP primer-based PCR, the five samples remained positive. No samples were PCR negative and EIA positive. The sensitivity, specificity, and positive and negative predictive values for the EIA compared with PCR were 85.3, 100, 100, and 77.5%, respectively. Table 1 shows that after retesting with MOMP primer, the plasmid primer-based PCR and MOMP primer-based PCR had detected equal numbers of chlamydia-positive samples. The sensitivity and specificity of PCR after retesting with MOMP primers were 100%.

**DISCUSSION**

To detect genital chlamydial infection in males, culture tests require urethral sampling, which asymptomatic persons are
PCR has been shown to be cost-effective for screening of adolescent males for *C. trachomatis* (10). Our study indicates that PCR is a suitable method for the detection of genital chlamydial infections in FUU samples from asymptomatic males.

**REFERENCES**


